

WHAT IS CLAIMED IS:

1. A vector containing at least following five DNA sequences: (1) a DNA sequence encoding one protein or its fragment; (2) a DNA sequence encoding a protein for displaying said one protein or its fragment on a phage; (3) a DNA sequence encoding another protein or its fragment; (4) a stop codon that enables display switch by a host strain; and (5) a DNA sequence encoding a protein for displaying said another protein or its fragment on the phage, the vector having a structure comprising these 5 DNA sequences in the order of (1), (2), (3), (4) and (5) or (3), (4), (5), (1) and (2) in the 5'-3' direction of the vector, by the presence of the stop codon that enables display switch by said host strain, when the vector is introduced into a suppressor-mutant host strain, the vector provides a two-protein displaying phage on which both of said one protein or its fragment and said another protein or its fragment are displayed, and when the vector is introduced into a non-suppressing host strain, the vector provides a one-protein displaying phage on which only said one protein or its fragment is displayed and said another protein or its fragment is secreted into the culture medium.

2. The vector as described in Claim 1 wherein said one protein or its fragment is a VH fragment of the variable region of an antibody, and said another protein or its fragment is a VL fragment of the variable region of an antibody.

3. The vector as described in Claim 1 wherein said one protein or its fragment is a VL fragment of the variable region of an antibody, and said another protein or its fragment is a VH fragment of the variable region of an antibody.

4. The vector as described in any one of Claims 1 to 3 wherein the vector is a phage vector or phagemid vector of E.coli.

5. The vector as described in any one of Claims 1 to 4 wherein said DNA sequence encoding a protein for displaying said one protein or its fragment on the phage is a DNA sequence encoding pIX protein

of a filamentous phage, and said DNA sequence encoding a protein for displaying said another protein or its fragment on the phage is a DNA sequence encoding pVII protein of a filamentous phage.

6. The vector as described in any one of Claims 1 to 4 wherein said DNA sequence encoding a protein for displaying said one protein or its fragment on the phage is a DNA sequence encoding pVII protein of a filamentous phage, and said DNA sequence encoding a protein for displaying said another protein or its fragment on the phage is a DNA sequence encoding pIX protein of a filamentous phage.

7. A vector as described in any one of Claims 1 to 6 wherein said stop codon that enables display switch by a host is an amber codon.

8. A method for determining interaction between one protein or its fragment and another protein or its fragment, the method comprising the steps of:

- (1) transforming a non-suppressing host strain by using the vector according any one of Claims 1 to 7, thereby obtaining one-protein displaying phage on which only said one protein or its fragment is displayed, and a culture medium containing said another protein or its fragment being secreted from said non-suppressing host strain;
- (2) immobilizing said another protein or its fragment in the supernatant on an appropriate support;
- (3) reacting said another protein or its fragment immobilized on the support with said one protein or its fragment displayed on the one-protein displaying phage, thereby said one protein or its fragment is bound to said another protein or its fragment; and
- (4) determining the amount of immobilized phages by an immunoassay using a labeled anti-phage antibody, thereby evaluating the binding ability between said one protein or its fragment and said another protein or its fragment.

9. The method as described in Claim 8 wherein said one protein or its fragment is a VH fragment of the variable region of an antibody, and said another protein or its fragment is a VL fragment of the

variable region of an antibody.

10. The method as described in Claim 8 wherein said one protein or its fragment is a VL fragment of the variable region of an antibody, and said another protein or its fragment is a VH fragment of the variable region of an antibody.

11. The method as described in any one of Claims 8 to 10 wherein the vector is a phage vector or phagemid vector of E.coli.

12. The method as described in any one of Claims 8 to 11 wherein said DNA sequence encoding a protein for displaying said one protein or its fragment on the phage is a DNA sequence encoding pIX protein of a filamentous phage, and said DNA sequence encoding a protein for displaying said another protein or its fragment on the phage is a DNA sequence encoding pVII protein of a filamentous phage.

13. The method as described in any one of Claims 8 to 11 wherein said DNA sequence encoding a protein for displaying said one protein or its fragment on the phage is a DNA sequence encoding pVII protein of a filamentous phage, and said DNA sequence encoding a protein for displaying said another protein or its fragment on the phage is a DNA sequence encoding pIX protein of a filamentous phage.

14. The method as described in any one of Claims 8 to 13 wherein said stop codon that enables display switch by a host is an amber codon.

15. The method as described in any one of Claims 8 to 14 wherein said suppressor-mutant host strain is an E.coli amber-suppressor strain.

16. The method as described in Claim 15 wherein said E.coli amber-suppressor strain is a strain TG1 of E. coli, and said non-suppressor host strain is E.coli strain HB2151.

17. A method for obtaining a variable region of an antibody in which interaction between a VH fragment and a VL fragment alters by the presence of an antigen, the method comprising the steps of:

(1) transforming a suppressor-mutant host strain by using a vector as described in Claim 2 to obtaining a VH/VL displaying phage, on

which both of the VH and VL fragments are displayed when said vector is introduced into the suppressor-mutant host strain;

(2) confirming the binding ability between the VH/VL displaying phage obtained in the step (1) and an antigen;

(3) transforming a non-suppressing host strain by using said vector as described in claim 2 whose ability to provide a VH/VL displaying phage is confirmed in the step (2), or transfecting a non-suppressing host strain with the VH/VL displaying phage obtained in the step (1), thereby obtaining a VH displaying phage on which only the VH fragment is displayed and a culture medium containing the VL fragment secreted by the non-suppressing host strain;

(4) immobilizing said VL fragment in the supernatant onto an appropriate support;

(5) reacting said VL fragment immobilized on the support with said VH fragment displayed on the phage in the presence or absence of an antigen;

(6) determining the amount of immobilized phages by an immunoassay using a labeled anti-phage antibody, thereby evaluating the binding ability between said VH fragment and said VL fragment; and

(7) determining that an antibody variable region in which interaction between the VH fragment and the VL fragment significantly alters in the presence of an antigen is obtained, provided that the binding ability between said VH fragment and said VL fragment in the presence of an antigen is two times or more than the binding ability between the VH fragment and the VL fragment in the absence of the antigen.

18. A method for obtaining a variable region of an antibody in which interaction between a VH fragment and a VL fragment alters by the presence of an antigen, the method comprising the steps of:

(1) transforming a suppressor-mutant host strain by using a vector as described in Claim 3 to obtaining a VH/VL displaying phage, on which both of the VH and VL fragments are displayed when said

vector is introduced into the suppressor-mutant host strain;

(2) confirming the binding ability between the VH/VL displaying phage obtained in the step (1) and an antigen;

(3) transforming a non-suppressing host strain by using said vector as described in claim 3 whose ability to provide a VH/VL displaying phage is confirmed in the step (2), or transfecting a non-suppressing host strain with the VH/VL displaying phage obtained in the step (1), thereby obtaining a VL displaying phage on which only the VL fragment is displayed and a culture medium containing the VH fragment secreted by the non-suppressing host strain;

(4) immobilizing said VH fragment in the supernatant onto an appropriate support;

(5) reacting said VH fragment immobilized on the support with said VL fragment displayed on the phage in the presence or absence of an antigen;

(6) determining the amount of immobilized phages by an immunoassay using a labeled anti-phage antibody, thereby evaluating the binding ability between said VH fragment and said VL fragment; and

(7) determining that an antibody variable region in which interaction between the VH fragment and the VL fragment significantly alters in the presence of an antigen is obtained, provided that the binding ability between said VH fragment and said VL fragment in the presence of an antigen is two times or more than the binding ability between the VH fragment and the VL fragment in the absence of the antigen.

19. A method for obtaining a VL fragment of the variable region of an antibody, the method comprising the steps of:

(1) transforming a non-suppressing host strain by using the vector as described in Claim 2, or transfecting a non-suppressing host strain with a VH/VL displaying phage containing said vector;

(2) allowing the transformed non-suppressing host strain to secrete the VL fragment into the culture medium; and

(3) purifying the VL fragment from the culture medium.

20. A method for obtaining a VH fragment of the variable region of an antibody, the method comprising the steps of:

(1) transforming a non-suppressing host strain by using a vector as described in Claim 3, or transfecting a non-suppressing host strain with a VH/VL displaying phage containing said vector;

(2) allowing the transformed non-suppressing host strain to secrete the VH fragment into the culture medium; and

(3) purifying the VH fragment from the culture medium.